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CENTRAL FAX CENTERAttorney Docket No. P69451US1
Application No. 10/586,329

OCT 05 2009

Amendments to the claims:

This listing of claims replaces all prior versions, and listings, of claims in the application.

Listing of claims:

Claim 1 (original): A method for examining the activity of ion channels, comprising the following steps:

- providing a sample comprising ion channels; and
- determining a value of a measuring parameter as an indicator of the activity of the ion channels, the measuring parameter being a membrane potential, a measure of a membrane potential, an ion concentration, or a measure of an ion concentration;

characterised in that said determining of the value of the measuring parameter is performed at a temperature of about 10 °C by fluorescence methods, radioactive methods or atomic absorption spectroscopy.

Claim 2 (original): The method according to claim 1, characterized in that said determining of the value of the measuring parameter is performed at a temperature of about 5 °C, especially about 2 °C.

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Claim 3 (previously presented): The method according to claim 1, characterized in that said determining of the value of the measuring parameter is performed at a temperature of from about 10 °C to 4 °C, especially from about 5 °C to -4 °C, more preferably from about 5 °C to 0 °C, even more preferably from about 2 °C to 0 °C.

Claim 4 (previously presented): The method according to claim 1, characterized in that the sample comprises one or more cells or cell organelles which have ion channels, in particular human or animal cells or cell organelles.

Claim 5 (previously presented): The method according to claim 1, characterized in that the sample comprises one or more vesicles which have ion channels.

Claim 6 (previously presented): The method according to claim 1, characterized in that the sample comprises membrane bound ion channels, in particular ion channels embedded into a membrane of cells, cell organelles, vesicles or embedded into an artificial membrane.

Claim 7 (previously presented): The method according to claim 1, characterized in that said measuring parameter is the membrane potential of a cell, cell organelle or vesicle, or a measure of said membrane potential.

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Claim 8 (previously presented): The method according to claim 1, characterized in that the measuring parameter is an extracellular, intracellular, extravesicular and/or intravesicular ion concentration or a measure thereof.

Claim 9 (previously presented): The method according to claim 1, characterized in that the value of said measuring parameter is determined before, during and/or after the addition of a test substance which potentially influences the activity of the ion channels.

Claim 10 (previously presented): The method according to claim 1, characterized in that the activity of a transmitter-dependent ion channel is examined.

Claim 11 (previously presented): The method according to claim 1, characterized in that the activity of a voltage-sensitive ion channel is examined.

Claim 12 (previously presented): The method according to claim 1, characterized in that the activity of a potassium channel, chloride channel, sodium channel or calcium channel is examined.

Claim 13 (previously presented): The method according to claim 1 characterized in that an optical response of (i) a carbocyanine derivative, in particular a thia-, indo-, or oxa-carbocyanine or an

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iodide derivative of a carbocyanine, (ii) a rhodamine dye, (iii) an oxonol dye, (iv) merocyanine 540, or (v) a styryl dye serves as a measure of the membrane potential.

Claim 14 (previously presented): The method according to claim 1, characterized in that the fluorescence emission of a voltage-sensitive fluorescent dye, preferably a DiBAC dye, more preferably the dye Dibac4(3), serves as a measure of the membrane potential.

Claim 15 (previously presented): The method according to claim 1, characterized in that the ion concentration of rubidium, especially of non-radioactive rubidium, is determined as an indicator of the activity of the ion channels.

Claim 16 (previously presented): The method according to claim 1, characterized in that the ion concentration, especially the ion concentration of calcium, is measured by means of chelating agents.

Claim 17 (previously presented): The method according to claim 1, characterized in that the values of several measuring parameters are determined.

Claim 18 (previously presented): The method according to claim 1 for use in the research on pharmaceutically active substances, especially in the medium- or high-throughput screening of potentially or established active pharmaceutical substances, in particular the identification of

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potentially active pharmaceutical substances or the determination of side effects of potentially or established active pharmaceutical substances.

Claim 19 (previously presented): The method according to claim 1 for use in the agricultural research, especially in the research on agrochemicals as e.g. insectizids.

Claim 20 (currently amended): ~~Use~~ A method of using a voltage-sensitive or ion-sensitive indicator ~~for the conductance of~~ in conducting the method according to claim 1.

Claim 21 (currently amended): ~~Use~~ The method according to claim 20 wherein the ion-sensitive indicator is a calcium indicator, in particular a fluo-calcium indicator, a fura indicator, an indo indicator, Calcium Green™, or Oregon Green™.

Claim 22 (currently amended): ~~Use~~ The method according to claim 20 wherein the ion-sensitive indicator is a sodium or potassium indicator, preferably a fluorescent sodium or potassium indicator, in particular SBFI, PBFI, Sodium Green Na⁺ indicator, CoroNa Green Na⁺ indicator, or CoroNa Red Na⁺ indicator.

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Claim 23 (currently amended): ~~Use~~ The method according to claim 20 wherein the voltage-sensitive indicator is a carbocyanine derivative, in particular an indo-, thia-, or oxa- carbocyanine or a iodide derivative of a carbocyanine; a rhodamine dye; an oxonol dye; merocyanine 540; or a styryl dye.

Claim 24 (currently amended): ~~Use~~ The method according to claim 23 wherein the oxonol dye is a bis-isoxazolone oxonol dye or a bis-barbituric acid oxonol (DiBAC) dye, in particular DiBAC4(3), DiSBAC2(3) or DiBAC4(5).

Claim 25 (currently amended): ~~Use~~ The method according to claim 23 wherein the styryl dye is an ANEP (AminoNaphthylEthenylPyridinium) dye, in particular di-4-ANEPPS, di-8-ANEPPS, di-2-ANEPEQ, di-8-ANEPPQ, di-12-ANEPPQ, di-1-ANEPIA, or a dialkylaminophenylpolyenylpyridinium dye (RH dye), in particular RH 414, RH 421, RH 795 or RH 237.

Claim 26 (currently amended): ~~Use~~ A method of using a chelating agent ~~for the conductance of in~~ conducting the method according to claim 1.

Claim 27 (currently amended): ~~Use~~ A method of using rubidium, in particular non-radioactive rubidium, ~~for the conductance of in conducting~~ the method according to claim 1.

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Claim 28 (currently amended): Use A method of using an atomic absorption spectrometer, a flow cytometer, a fluorescence microscope or fluorescence plate reader ~~for the conductance of in~~ conducting the method according to claim 1.

Claim 29 (currently amended): Use A method of using an atomic absorption spectrometer, a flow cytometer, a fluorescence microscope or fluorescence plate reader for applying a voltage-sensitive or ion-sensitive indicator according to claim 20 .

Claim 30 (currently amended): Use A method of using an atomic absorption spectrometer, a flow cytometer, a fluorescence microscope or fluorescence plate reader for applying a chelating agent according to claim 26.

Claim 31 (currently amended): Use A method of using an atomic absorption spectrometer, a flow cytometer, a fluorescence microscope or fluorescence plate reader for applying rubidium according to claim 27.